

January 23, 1947.

Dear Dr. Braun,

I was happy to hear from you that you are continuing with the genetic aspects of the nature of your *Phytomonas* forms. Prof. Tatum and I have developed a simplified technique for the detection of biochemical mutants of bacteria, which works quite well with some organisms. A reprint is enclosed. If *Phytomonas* will form regular colonies in minimal agar medium, with of satisfactory size, and little variation in size, this technique should work very well in screening large samples of irradiated bacteria to find mutants. Otherwise, the older techniques described by Gray and Tatum, etc., would be preferable. The only possible quirk that must be mentioned is that is probably advisable to incubate the irradiated cultures for several hours in fresh medium before plating them out. While we have not yet completed critical studies on this point, this should be necessary to allow the possible segregation of mutated from un-mutated nuclei in what appears to be a di-karyotic cell. With material such as yours, however, where the occurrence of star forms is apparently frequent under certain conditions, it may not be necessary to rely on biochemical mutants. Dr. Burkholder, in this laboratory, spent considerable time last summer in looking for mutants in *Phytomonas*, without success. This may have been because the importance of the incubation period was not fully understood.

The rarity of the recombination process in *E. coli* required us to use mutant characters ~~which~~ for the recombinations of which we could select by manipulation of the medium. Since that time, we have been using nutritional characters as a means of selecting for the recombination cells, but have used other markers which can be more conveniently scored (such as colony form, lactose fermentation, virus resistance, etc.) On the hypothesis that the stars represent or contain zygotes, almost any reasonably stable mutant character should be adequate; the occurrence of recombinations of such characters (plated in toto) in cells of colonies obtained from stars, and not otherwise would be critical evidence. Such mutants should be readily picked up to judge from the work of Lincoln and Gowen on *P. stewartii*. Another type of character that can be obtained very readily is drug resistance: several drugs are available which will select for mutant bacteria resistant to them, and do not show cross-resistance, e.g. penicillin and streptomycin.

At any rate, the best of luck to you, and let us hear what comes out.

Yours sincerely,


Joshua Lederberg.)